linear concentration gradient, which leads to the parabolic law, the concentration of diffusion species would decrease exponentially along the diffusion paths, or

#### $c = c_0 e^{-\alpha x}$

where c is the concentration of the diffusion species at a distance x from its starting point. Thus, the concentration gradient would be

$$\partial c/\partial x = -c_0 k e^{-\alpha}$$

Since the total thickness of oxide, x, is related to

the amount of substance that has diffused, w, by a gravimetric factor w = gx, then

 $w = k \log(1 + at)$ 

 $\mathrm{d}w/\mathrm{d}t = -Dc_0\alpha e^{-w\alpha/g}$ 

Integrating

which is the logarithmic law for oxidation.

**Acknowledgment**.—The authors are grateful to the office of Naval Research for sponsoring this work.

CHICAGO 16, ILL. RECEIVED NOVEMBER 20, 1950

# NOTES

### Quaternary Salts of Halogenated Pyridines and Quinolines<sup>1</sup>

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Since certain quaternary salts of pyridine and quinoline have been reported to damage sarcoma cells *in vivo*<sup>2</sup> we have prepared similar salts of several halogenated pyridines and quinolines for screening against sarcoma in mice<sup>3</sup> and for correlation of biological effects and other properties with structure.

The quaternary salts listed in Tables I and II were prepared by reaction of a halogenated heterocyclic base with the appropriate organic halide at  $30-40^{\circ}$ . When the reactants alone did not form a homogeneous solution a small amount of chloroform was added to bring them into solution. The products usually precipitated as they were formed

# Table I

#### HALOPYRIDINE DERIVATIVES

Empirical	M.p.,	Analyses, % Ionic Halogen	
101 111112	C	Calco.	roundo
L <sub>13</sub> H <sub>18</sub> BrClN	193	26.76	26.50
L13H18BrClNO	182	25.40	25.30
L <sub>13</sub> H <sub>11</sub> BrClNO	187	25.57	25.30
L <sub>17</sub> H <sub>19</sub> BrClNO	192	21.68	21.40
13H10BrClFNO	185–1 <b>87</b>	24.17	24.03
L13H10BrCl2NO	188-189	23.03	23.01
$L_{13}H_{10}Br_2C1NO$	194	20.41	20.42
13H10BrClINO	193	18.23	18.00
$_{13}H_{10}BrClN_2O_3$	172	22.35	22.42
13H10Br2FNO	174-175	21.30	21.08
$L_{13}H_{10}Br_2C1NO$	201	20.41	20.44
<sub>13</sub> H <sub>10</sub> Br <sub>2</sub> INO	189	16.55	16.40
19H15Br2NO	160-161	18.45	18.54
$_{17}H_{17}Br_2NO$	207	19.44	19.54
$_{13}\mathrm{H_{10}BrF_2NO}$	189	25.36	25.29
13H10BrClNO	169-170	24.17	24.33
	Empirical formula 213H18BrClN 213H18BrClNO 213H11BrClNO 213H10BrClNO 213H10BrClPNO 213H10BrCl2NO 213H10Br2ClNO 213H10Br2ClNO 213H10Br2ClNO 213H10Br2CNO 213H10Br2PNO 213H10Br2NO 213H10Br2NO 213H10BrF2NO 213H10BrF2NO 213H10BrF2NO 213H10BrF2NO 213H10BrF2NO 213H10BrF2NO 213H10BrF2NO	Empirical formula         M.p., °C.a $h_{14}H_{14}BrClN$ 193 $h_{14}H_{14}BrClNO$ 182 $h_{14}H_{14}BrClNO$ 187 $h_{14}H_{14}BrClNO$ 192 $h_{14}H_{16}BrClNO$ 187 $h_{14}H_{10}BrClNO$ 192 $h_{14}H_{10}BrClNO$ 185–187 $h_{14}H_{10}BrCl_2NO$ 188–189 $h_{14}H_{10}BrClNO$ 193 $h_{14}H_{10}Br_{2}ClNO$ 193 $h_{14}H_{10}Br_{2}FNO$ 174–175 $h_{14}H_{10}Br_{2}ClNO$ 201 $h_{14}H_{10}Br_{2}NO$ 160–161 $h_{17}H_{17}Br_{2}NO$ 207 $h_{14}H_{10}BrF_{2}NO$ 189 $h_{14}H_{10}BrF_{2}NO$ 189 $h_{14}H_{10}BrF_{2}NO$ 189	Empirical formulaM.p., °C.aTonic Hall Tonic Hall Calcd. $h_{13}H_{14}BrClN19326.76h_{14}H_{13}BrClNO18225.40h_{14}H_{11}BrClNO18725.57h_{17}H_{19}BrClNO19221.68h_{13}H_{10}BrClFNO185-18724.17h_{13}H_{10}BrClNO19420.41h_{14}H_{10}Br_2ClNO19318.23h_{13}H_{10}Br_2ClNO19420.41h_{14}H_{10}Br_2ClNO19318.23h_{13}H_{10}Br_2ClNO20120.41h_{14}H_{10}Br_2ClNO20120.41h_{14}H_{10}Br_2ClNO20120.41h_{13}H_{10}Br_2CNO160-16118.45h_{17}H_{17}Br_2NO20719.44h_{14}H_{10}BrF_2NO18925.36h_{14}H_{10}BrClNO169-17024.17$

(1) This investigation was supported in part by a research grant from the National Cancer Institute, of the National Institutes of Health, Public Health Service. and the mixture was allowed to stand as long as seemed necessary to obtain a good yield. The rates of reaction varied greatly. For 6-chloroquinoline the reaction periods were: with glycerol-(3) Results of screening tests at the National Cancer Institute are to be reported elsewhere.

<sup>(2)</sup> Shear, et al., in "Approaches to Cancer Chemotherapy," American Association for the Advancement of Science, F. R. Moulton, Editor, Washington, D. C., 1947, p. 236 ff.; cf. J. L. Hartwell and S. R. L. Kornberg, THIS JOURNAL, 69, 1131 (1946).

## Notes

$\begin{array}{c c c c c c c c c c c c c c c c c c c $	TABLE I   (Continued)							
$\begin{array}{rrrrrrrrrrrrrrrrrrrrrrrrrrrrrrrrrrrr$		Empirical formula	$\mathbf{M}_{\mathbf{p}_{a}}$	Analy Ionic I Caled.	7ses, % Halogen Foundb			
$\begin{array}{llllllllllllllllllllllllllllllllllll$	3-Bromopyridine and			~				
2,5-Diodohexane (bis-sait) $C_{18}H_{20}H_{2} = 2445 = 38.93 = 38.83$ $Elly erol a, -p-dibromohydrin C_{11}H_{11}Br_1N_0 = 330 = 29.83Ethyl iodoacetate C_{11}H_{11}Br_1N_0 = 178-179 = 34.12 = 34.53Cyclohexylethyl bromide C_{11}H_{13}Br_2NO = 216-217 = 22.52 = 29.06Eyrene bromohydrin C_{11}H_{13}Br_2NO = 210-211 = 19.34 = 19.642,5-Dimethylphenacyl bromide C_{12}H_{13}Br_2NO = 210-211 = 19.34 = 19.642,5-Dimethylphenacyl bromide C_{12}H_{13}Br_2NO = 210-211 = 21.28 = 20.97p-Chlorophenacyl bromide C_{12}H_{13}Br_1NO = 238-230 = 16.55 = 16.572,5-Dichlorophenacyl bromide C_{13}H_{16}Br_2(DN = 238-230 = 18.76 = 18.74m-Nitrophenacyl bromide C_{13}H_{14}Br_2(LNO = 238-230 = 18.76 = 18.74$ $m$ -Nitrophenacyl bromide C_{14}H_{13}Br_1NO = 243 = 20.64 = 20.76 = 20.64 = 20.	Decyl iodide	$C_{15}H_{25}BrIN$	80	29.77	29.92			
$\begin{array}{llllllllllllllllllllllllllllllllllll$	2,5-Diiodohexane (bis-salt)	$C_{16}H_{20}I_4N_2$	244-245	38.93	38.83			
Ethyl iodoacetate       C, H <sub>11</sub> Br1N0,       178       179       34.12       34.53         Cyclohexylethyl bromide       C <sub>11</sub> H <sub>13</sub> Br <sub>2</sub> N       123-125       22.96         Styrene bromohydrin       C <sub>14</sub> H <sub>13</sub> Br <sub>2</sub> NO       216-217       22.25       22.96         Styrene bromohydrin       C <sub>14</sub> H <sub>13</sub> Br <sub>2</sub> NO       216-217       22.25       22.51         β-Fluorophenacyl bromide       C <sub>14</sub> H <sub>13</sub> Br <sub>2</sub> NO       254       20.75       20.73         β-Fluorophenacyl bromide       C <sub>14</sub> H <sub>13</sub> Br <sub>2</sub> CINO       238-240       20.40       20.41         β-Iodophenacyl bromide       C <sub>14</sub> H <sub>13</sub> Br <sub>2</sub> CINO       238-239       18.76       18.74         m-Nitrophenacyl bromide       C <sub>14</sub> H <sub>13</sub> Br <sub>12</sub> NO <sub>3</sub> 207-209       19.87       19.73         3,4-Dihydroxyphenacyl bromide       C <sub>14</sub> H <sub>13</sub> Br <sub>12</sub> NO       243       20.64       20.64         β-Naphthacyl bromide       C <sub>14</sub> H <sub>13</sub> Br <sub>12</sub> NO       242-235       19.63       19.29         β-Naphthacyl bromide       C <sub>14</sub> H <sub>13</sub> Br <sub>12</sub> NO       242-235       19.63       19.29         β-Naphthacyl bromide       C <sub>14</sub> H <sub>13</sub> Br <sub>12</sub> NO       242-235       19.63       19.29         β-Naphthacyl bromide       C <sub>14</sub> H <sub>13</sub> Br <sub>12</sub> NO       24-215       27.76       27.76         anti-β-Nap	Glycerol- $\alpha$ , $\gamma$ -dibromohydrin	$C_{13}H_{14}Br_4N_2O$	330	29.93	29.83			
$ \begin{array}{llllllllllllllllllllllllllllllllllll$	Ethyl iodoacetate	$C_9H_{11}BrINO_2$	178-179	34.12	34.53			
Styrene bromohydrinCi <sub>1</sub> H <sub>10</sub> Br <sub>2</sub> NO216-21722.2522.51 $\rho$ -Futylphenacyl bromideCi <sub>1</sub> H <sub>10</sub> Br <sub>2</sub> NO210-21119.3419.64 $\rho$ -Fluorophenacyl bromideCi <sub>1</sub> H <sub>10</sub> Br <sub>2</sub> NO25420.7520.73 $\rho$ -Fluorophenacyl bromideCi <sub>1</sub> H <sub>10</sub> Br <sub>2</sub> NO25420.7520.73 $\rho$ -Chlorophenacyl bromideCi <sub>1</sub> H <sub>10</sub> Br <sub>2</sub> NO268-24020.4020.41 $\rho$ -Iodophenacyl bromideCi <sub>1</sub> H <sub>10</sub> Br <sub>2</sub> NO238-23918.7618.76 $\rho$ -Diodophenacyl bromideCi <sub>1</sub> H <sub>10</sub> Br <sub>2</sub> NO238-23918.7618.74 $m$ -Nitrophenacyl bromideCi <sub>1</sub> H <sub>10</sub> Br <sub>1</sub> NO238-23918.7618.74 $m$ -Nitrophenacyl bromideCi <sub>1</sub> H <sub>10</sub> Br <sub>1</sub> NO24320.6420.64 $\rho$ -Methoxyphenacyl bromideCi <sub>1</sub> H <sub>10</sub> Br <sub>1</sub> NO214-21527.9527.76 $\rho$ -Mathoxyl bromideCirH <sub>10</sub> Br <sub>1</sub> NO214-21527.9527.76 $\rho$ -Naphthacyl bromideCirH <sub>10</sub> Br <sub>1</sub> NO214-21527.9527.76 $\rho$ -Raphthacyl bromideCirH <sub>10</sub> Br <sub>1</sub> NO214-21527.9527.76 $\sigma$ -PropionaphthoneCi <sub>1</sub> H <sub>10</sub> Br <sub>2</sub> NO220-22119.4419.24 $\phi$ -Bromo- $\rho$ -propionaphthoneCi <sub>10</sub> H <sub>10</sub> Br <sub>2</sub> NO220-22119.4419.57 $\phi$ -Dioophenacyl bromideCirH <sub>10</sub> Br <sub>2</sub> NO220-22119.4419.57 $\phi$ -Dioophenacyl bromideCi <sub>10</sub> H <sub>10</sub> Br <sub>2</sub> NO220-22119.4419.57 $\phi$ -Dioophenacyl bromideCirH <sub>10</sub> Br <sub>2</sub> NO220-22119.7119.57 $\phi$ -Dioophenacyl bromide	Cyclohexylethyl bromide	$C_{13}H_{19}Br_2N$	123 - 125	22.90	22.96			
$\begin{array}{cccccccccccccccccccccccccccccccccccc$	Styrene bromohydrin	$C_{13}H_{13}Br_2NO$	216-217	22.25	22.51			
2,5-Dimethylphenacyl bromide $C_{18}H_{18}Br_{2}NO$ 254 20,75 20.73 $p$ -Fluorophenacyl bromide $C_{18}H_{19}Br_{2}FNO$ 112 21.28 20.97 $p$ -Chlorophenacyl bromide $C_{18}H_{19}Br_{2}FNO$ 236–240 20.40 20.41 $p$ -Iodophenacyl bromide $C_{18}H_{19}Br_{2}INO$ 268–270 16.55 16.57 2,5-Dichlorophenacyl bromide $C_{18}H_{19}Br_{2}LNO$ 238–239 18.76 18.74 $m$ -Nitrophenacyl bromide $C_{18}H_{19}Br_{2}NO$ 207–209 19.87 19.73 $3$ ,4-Dihydroxyphenacyl bromide $C_{18}H_{19}Br_{2}NO$ 243 20.64 20.64 $\beta$ -Naphthacyl bromide $C_{17}H_{18}Br_{2}NO$ 234–235 19.63 19.29 $\beta$ -Naphthacyl iodide $C_{17}H_{18}Br_{1}NO$ 214–215 27.95 27.76 $amti-\beta$ -Naphthacyl iodide $C_{17}H_{18}Br_{1}NO$ 202–203 $d$ $4$ -Fluoro- $\alpha$ -naphthacyl bromide $C_{17}H_{18}Br_{2}NO$ 202–203 $d$ $4$ -Fluoro- $\alpha$ -naphthacyl bromide $C_{17}H_{18}Br_{2}NO$ 202–203 $d$ $4$ -Fluoro- $\alpha$ -naphthacyl bromide $C_{17}H_{18}Br_{2}NO$ 202–221 19.44 19.24 $\alpha$ -Bromo- $\beta$ -propionaphthone $C_{18}H_{18}Br_{2}NO$ 203–204 19.71 19.57 3-Iodopyridine and $p$ -Chlorophenacyl bromide $C_{16}H_{48}Br_{4}NO$ 203–204 19.71 19.57 3-Iodopyridine and Decyl iodide $C_{17}H_{18}Br_{1}NO$ 202–204 18.98 19.08 3,5-Dibromopyridine and Decyl iodide $C_{16}H_{48}Br_{4}NO$ 190–191 16.24 15.93 $p$ -Fluorophenacyl bromide $C_{16}H_{48}Br_{5}NO$ 220 17.60 17.36 $p$ -Fluorophenacyl bromide $C_{17}H_{18}Br_{5}NO$ 220 17.60 17.36 $p$ -Chlorophenacyl bromide $C_{16}H_{48}Fr_{5}NO$ 220 17.60 17.36 p	<i>p-t</i> -Butylphenacyl bromide	$C_{17}H_{19}Br_2NO$	210-211	19.34	19.64			
$\begin{array}{llllllllllllllllllllllllllllllllllll$	2,5-Dimethylphenacyl bromide	$C_{15}H_{15}Br_2NO$	254	20.75	20.73			
$\begin{array}{llllllllllllllllllllllllllllllllllll$	<i>p</i> -Fluorophenacyl bromide	$C_{13}H_{10}Br_2FNO$	112	21.28	20.97			
	<i>p</i> -Chlorophenacyl bromide	$C_{13}H_{10}Br_2CINO$	236 - 240	20.40	20.41			
$\begin{array}{llllllllllllllllllllllllllllllllllll$	<i>p</i> -Iodophenacyl bromide	$C_{13}H_{10}Br_2INO$	268 - 270	16.55	16.57			
$\begin{array}{cccccccccccccccccccccccccccccccccccc$	2,5-Dichlorophenacyl bromide	$C_{13}H_9Br_2Cl_2NO$	238 - 239	18.76	18.74			
$\begin{array}{rrrrrrrrrrrrrrrrrrrrrrrrrrrrrrrrrrrr$	<i>m</i> -Nitrophenacyl bromide	$C_{13}H_{10}Br_2N_2O_3$	207 - 209	19.87	19.73			
$\begin{array}{cccccccccccccccccccccccccccccccccccc$	3,4-Dihydroxyphenacyl bromide	C <sub>18</sub> H <sub>11</sub> BrClNO <sub>3</sub>	252	1	D			
$\begin{array}{llllllllllllllllllllllllllllllllllll$	<i>p</i> -Methoxyphenacyl bromide	$C_{14}H_{13}Br_2NO_2$	243	20.64	20.64			
$\begin{array}{cccccccccccccccccccccccccccccccccccc$	$\beta$ -Naphthacyl bromide	$C_{17}H_{13}Br_2NO$	234 - 235	19.63	19.29			
anti- $\beta$ -Naphthacyl iodide oxime $C_{17}H_{14}BrIN_2O$ $202-203$ $d$ 4-Fluoro- $\alpha$ -naphthacyl bromide $C_{17}H_{12}Br_2FNO$ $214$ $18.80$ $18.52$ 5,6,7,8-Tetrahydro- $\beta$ -naphthacyl bromide $C_{17}H_{12}Br_2NO$ $220-221$ $19.44$ $19.24$ $\alpha$ -Bromo- $\beta$ -propionaphthone $C_{18}H_{13}Br_2NO$ $234-235$ $18.98$ $18.83$ $p$ -Chlorophenyl- $\alpha$ -bromoethyl ketone $C_{18}H_{20}I_4N_2$ $261-264$ $34.02$ $34.03$ $p$ -Fluorophenacyl bromide $C_{16}H_{20}I_4N_2$ $261-264$ $34.02$ $34.03$ $p$ -Fluorophenacyl bromide $C_{16}H_{20}I_4N_2$ $261-264$ $34.02$ $34.03$ $p$ -Fluorophenacyl bromide $C_{16}H_{20}I_4N_2$ $261-264$ $34.02$ $34.03$ $p$ -Fluorophenacyl bromide $C_{16}H_{24}Br_2IN$ $208-209$ $15.82$ $15.95$ $\beta$ -Phenylethyl iodide $C_{17}H_{13}Br_3NO$ $190-191$ $16.24$ $15.93$ $p$ -Fluorophenacyl bromide $C_{13}H_{4}Br_3FNO$ $220$ $17.60$ $17.36$ $p$ -Fluorophenacyl bromide $C_{13}H_{3}Br_3NO$ $225-226$ $16.98$ $17.21$ $p$ -Bromophenacyl bromide $C_{13}H_9Br_8INO$ $237$ $14.22$ $14.48$ $m$ -Nitrophenacyl bromide $C_{13}H_9Br_8NO_2$ $237$ $14.22$ $14.48$ $p$ -Methoxyphenacyl bromide $C_{13}H_9Br_8NO_2$ $2351$ $17.15$ $17.15$ $p$ -Chlorophenyl $\alpha$ -bromotehyl ketone $C_{14}H_{12}Br_8NO_2$ $251$ $17.15$ $17.15$ $p$ -Didophenacyl bromide $C_{12}H_{12}Br_8NO_2$ </td <td><math>\beta</math>-Naphthacyl iodide</td> <td>C<sub>17</sub>H<sub>13</sub>BrINO</td> <td>214 - 215</td> <td>27.95</td> <td>27.76</td>	$\beta$ -Naphthacyl iodide	C <sub>17</sub> H <sub>13</sub> BrINO	214 - 215	27.95	27.76			
$\begin{array}{llllllllllllllllllllllllllllllllllll$	anti-β-Naphthacyl iodide oxime	$C_{17}H_{14}BrIN_2O$	202 - 203		đ			
$\begin{array}{cccccccccccccccccccccccccccccccccccc$	4-Fluoro-α-naphthacyl bromide	$C_{17}H_{12}Br_2FNO$	214	18.80	18.52			
$\begin{array}{cccccccccccccccccccccccccccccccccccc$	5,6,7,8-Tetrahydro-β-naphthacyl bromide	$C_{17}H_{17}Br_2NO$	220-221	19.44	19.24			
$\begin{array}{cccccccccccccccccccccccccccccccccccc$	$\alpha$ -Bromo- $\beta$ -propionaphthone	$C_{18}H_{15}Br_2NO$	234 - 235	18.98	18.83			
$\begin{array}{c c c c c c c c c c c c c c c c c c c $	$p$ -Chlorophenyl- $\alpha$ -bromoethyl ketone	$C_{14}H_{12}Br_2C1NO$	203 - 204	19.71	19.57			
$\begin{array}{cccccccccccccccccccccccccccccccccccc$	3-Iodopyridine and							
$\begin{array}{cccccccccccccccccccccccccccccccccccc$	2,5-Diiodohexane	$C_{16}H_{20}I_4N_2$	261 - 264	34.02	34.03			
3,5-Dibromopyridine andDecyl iodide $C_{16}H_{24}Br_2IN$ $208-209$ $15.82$ $15.95$ $\beta$ -Phenylethyl iodide $C_{13}H_{13}Br_2IN$ $206-207$ $27.01$ $27.13$ $p$ -t-Butylphenacyl bromide $C_{17}H_{18}Br_3NO$ $190-191$ $16.24$ $15.93$ $p$ -Fluorophenacyl bromide $C_{13}H_9Br_3FNO$ $220$ $17.60$ $17.36$ $p$ -Chlorophenacyl bromide $C_{13}H_9Br_3CINO$ $225-226$ $16.98$ $17.21$ $p$ -Bromophenacyl bromide $C_{13}H_9Br_4NO$ $227-228$ $15.52$ $15.64$ $p$ -Iodophenacyl bromide $C_{13}H_9Br_8INO$ $237$ $14.22$ $14.48$ $m$ -Nitrophenacyl bromide $C_{13}H_9Br_8N_2O_3$ $238$ $16.62$ $16.40$ $p$ -Methoxyphenacyl bromide $C_{14}H_{12}Br_3NO_2$ $251$ $17.15$ $17.15$ $p$ -Chlorophenyl- $\alpha$ -bromoethyl ketone $C_{14}H_{11}Br_3CINO$ $192$ $16.50$ $16.51$ $p$ -Phenylphenacyl bromide $C_{19}H_{14}Br_3NO$ $216-217$ $15.60$ $15.80$	<i>p</i> -Fluorophenacyl bromide	$C_{13}H_{10}BrFINO$	202 - 204	18.98	19.08			
$\begin{array}{llllllllllllllllllllllllllllllllllll$	3.5-Dibromopyridine and							
$\begin{array}{llllllllllllllllllllllllllllllllllll$	Decyl iodide	$C_{15}H_{24}Br_2IN$	208 - 209	15.82	15.95			
$\begin{array}{llllllllllllllllllllllllllllllllllll$	$\beta$ -Phenylethyl iodide	$C_{13}H_{13}Br_2IN$	206 - 207	27.01	27.13			
$\begin{array}{llllllllllllllllllllllllllllllllllll$	<i>p-t</i> -Butylphenacyl bromide	C <sub>17</sub> H <sub>18</sub> Br <sub>3</sub> NO	190-191	16.24	15.93			
$\begin{array}{llllllllllllllllllllllllllllllllllll$	<i>p</i> -Fluorophenacyl bromide	C <sub>13</sub> H <sub>9</sub> Br <sub>3</sub> FNO	220	17.60	17.36			
$ \begin{array}{llllllllllllllllllllllllllllllllllll$	<i>p</i> -Chlorophenacyl bromide	C13H9Br3C1NO	225 - 226	16.98	17.21			
$ \begin{array}{cccccccccccccccccccccccccccccccccccc$	<i>p</i> -Bromophenacyl bromide	$C_{13}H_9Br_4NO$	227 - 228	15.52	15.64			
$ \begin{array}{llllllllllllllllllllllllllllllllllll$	<i>p</i> -Iodophenacyl bromide	C <sub>13</sub> H <sub>9</sub> Br <sub>3</sub> INO	237	14.22	14.48			
$ \begin{array}{llllllllllllllllllllllllllllllllllll$	<i>m</i> -Nitrophenacyl bromide	$C_{13}H_9Br_3N_2O_3$	238	16.62	16.40			
p-Chlorophenyl- $\alpha$ -bromoethyl ketone         C <sub>14</sub> H <sub>11</sub> Br <sub>3</sub> ClNO         192         16.50         16.51           p-Phenylphenacyl bromide         C <sub>19</sub> H <sub>14</sub> Br <sub>3</sub> NO         216-217         15.60         15.80	p-Methoxyphenacyl bromide	$C_{14}H_{12}Br_3NO_2$	251	17.15	17.15			
<i>p</i> -Phenylphenacyl bromide C <sub>19</sub> H <sub>14</sub> Br <sub>3</sub> NO 216-217 15.60 15.80	$p$ -Chlorophenyl- $\alpha$ -bromoethyl ketone	C14H11Br3CINO	192	16.50	16.51			
• • • • •	p-Phenylphenacyl bromide	C <sub>19</sub> H <sub>14</sub> Br <sub>3</sub> NO	216 - 217	15.60	15.80			
$\beta$ -Naphthacyl bromide $C_{17}H_{12}Br_3NO$ 203–204 16.44 16.19	β-Naphthacyl bromide	C <sub>17</sub> H <sub>12</sub> Br <sub>3</sub> NO	203 - 204	16.44	16.19			
$\beta$ -Naphthacyl iodide $C_{17}H_{12}Br_{3}INO$ 180–181 23.81 23.99	β-Naphthacyl iodide	C <sub>17</sub> H <sub>12</sub> Br <sub>2</sub> INO	180-181	23.81	23.99			
5,6,7,8-Tetrahydro- $\beta$ -naphthacyl bromide $C_{17}H_{18}Br_3NO$ 231–232 16.30 16.40	5.6.7.8-Tetrahydro- $\beta$ -naphthacvl bromide	C <sub>17</sub> H <sub>16</sub> Br <sub>3</sub> NO	231-232	16.30	16.40			
5,6,7,8-Tetrahydro- $\beta$ -naphthacyl iodide $C_{17}H_{18}Br_2INO$ 205 23.62 23.78	5,6,7,8-Tetrahydro- $\beta$ -naphthacyl iodide	C <sub>17</sub> H <sub>16</sub> Br <sub>2</sub> INO	205	23.62	23.78			

<sup>a</sup> Salts melted with decomposition. <sup>b</sup> Average of two Volhard analyses, unless otherwise indicated. <sup>c</sup> Calcd.: C, 45.32; H, 3.22. Found: C, 45.26; H, 3.48. <sup>d</sup> Calcd.: C, 43.53; H, 3.01. Found: C, 43.46; H, 3.12.

 $\alpha, \gamma$ -dibromohydrin<sup>4</sup> 60 days, with decyl iodide<sup>4</sup> 14 days, with  $\beta$ -phenylethyl bromide<sup>4</sup> 18 days, with  $\beta$ -phenylethyl iodide<sup>4</sup> 18 hours, with phenacyl bromide<sup>5</sup> 14 days, with p-methoxyphenacyl bromide<sup>5</sup> 4 days, with p-iodophenacyl bromide<sup>5</sup> 24 hours and with  $\beta$ -naphthacyl bromide<sup>5</sup> 24 hours. Among the bases, 3,5-dibromopyridine and 3-bromopyridine reacted more rapidly than 2-chloropyridine and 2-bromopyridine, while 4,7-dichloroquinoline reacted much less rapidly than 6-chloroquinoline. The results observed were in line with the expected deactivating effect of a negative atom attached at the 2- or 4- position on the heterocyclic ring and the steric hindering by a large atom or group attached to the carbon adjacent to the nitrogen.

The bromide salts were white or cream solids

(4) Without solvent.

(5) In chloroform.

while the iodides were yellow. Some of those with low molecular weights were very soluble in water, while others were only slightly soluble. Most of the salts were recrystallized by dissolving in warm methanol, ethanol or ethyl acetate and adding isopropyl ether, but some were recrystallized from water, alcohol or acetone without the aid of isopropyl ether.

Acknowledgments.—The authors wish to express their appreciation to Dr. M. J. Shear and Dr. J. L. Hartwell for arranging screening tests against mouse tumors and securing carbon and hydrogen analyses on some of the compounds, to Miss Marguerite Close for part of the Volhard analyses, to Mr. Hugh Jenkins, Mr. Clifford Myers, Mr. Jack Brasher, Mr. Gene Moore and Mr. Paul Scott for preparation of some of the organic halides used, to Dr. Arthur Roe for

#### Notes

## TABLE II HALOQUINOLINE DERIVATIVES

Salt from 6-chloroquinoline and	Empirical	M.n.	Analyses, % Ionic halogen	
	formula	°Ċ.	Calcd.	Found
Decyl iodide	C <sub>19</sub> H <sub>27</sub> ClIN	113	29.39	29.33
Glycerol- $\alpha$ , $\gamma$ -dibromohydrin	$C_{12}H_{12}Br_2ClNO$	234	20.95	21.22
$\beta$ -Cyclohexylethyl bromide	$C_{17}H_{21}BrClN$	102	22.52	22.33
$\beta$ -Phenylethyl bromide	C <sub>17</sub> H <sub>15</sub> BrClN	108-111	22.92	22.83
$\beta$ -Phenylethyl iodide	C <sub>17</sub> H <sub>15</sub> CIIN	164	32.09	31.90
Phenacyl bromide	C17H13Br CINO	215	22.04	21.78
<i>p-t</i> -Butylphenacyl bromide	$C_{21}H_{21}BrClNO$	232	19.08	18.98
<i>p</i> -Chlorophenacyl bromide	$C_{17}H_{12}BrCl_2NO$	205	20.12	20.12
<i>p</i> -Bromophenacyl bromide	$C_{17}H_{12}Br_2CINO$	207	18.09	18.11
<i>m</i> -Nitrophenacyl bromide	$C_{17}H_{12}BrClNO_3$	215	19.60	19.52
<i>p</i> -Methoxyphenacyl bromide	C <sub>18</sub> H <sub>15</sub> BrClNO	211	20.34	30.33
$\beta$ -Naphthacyl bromide	$C_{21}H_{15}BrClNO$	236.5	19.37	$19 \ 43$
5,6,7,8-Tetrahydro- $\beta$ -naphthacyl bromide	$C_{21}H_{19}BrClNO$	252	19.17	18.95
3-Bromoquinoline and				
<i>p</i> -Fluorophenacyl bromide	$C_{17}H_{12}Br_2FNO$	258	18.80	18.84

samples of 3-fluoro, 3-chloro and 3-iodopyridine, and to Miss Emogene Stephen, Miss Carolyn Cate, Mr. Tom Fuller and Mr. Lynn Easley for assistance in the purification of the products.

DEPARTMENT OF CHEMISTRY

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JEFFERSON CITY, TENN. **Received February 21, 1951** 

### Color Reactions of Human Antibody and Normal Human Gamma Globulin<sup>1</sup>

#### BY SAM M. BEISER AND ELVIN A. KABAT

As a criterion of purity of the blood group A substances<sup>2,3,4,5</sup> determinations were carried out of the proportions of two characteristic constituents of these antigens, hexosamine and methylpentose, specifically precipitated by an excess of antibodies to A substance. Since such specific precipitates consist of both antigen and antibody, total color values for hexosamine6 and methylpentose7 in specific precipitates must be corrected for any color given in these reactions by the antibody. The equivalent color values of normal human  $\gamma$ -globulin were used for this purpose although the possibility was recognized<sup>2</sup> and commented upon<sup>8</sup> that human antibody and normal human gamma globulin may not give identical color values. This report shows that human antipneumococcal antibodies give color values identical with those for human gamma globulin in the reactions for hexosamine and methylpentose as well as with Folin-Ciocalteu tyrosine reagent.9

(1) The work reported in this paper was carried out under a research grant from the Division of Research Grants and Fellowships of the National Institutes of Health, United States Public Health Service and in part under the William J. Matheson Commission.

(2) A. Bendich, E. A. Kabat and A. E. Bezer, J. Exp. Med., 83, 485 (1946).

(3) E. A. Kabat, A. Bendich, A. E. Bezer and S. M. Beiser. ibid., 85, 685 (1947).

(4) E. A. Kabat, H. Baer and V. Knaub, ibid., 89, 1 (1949).

(5) E. A. Kabat, Bact. Revs., 13, 189 (1949).

(6) L. A. Elson and W. T. J. Morgan, Biochem. J., 27, 1824 (1933).

(7) Z. Dische and L. B. Shettles, J. Biol. Chem., 175, 595 (1948).

(8) G. Holzman and C. Niemann, THIS JOURNAL, 72, 2048 (1950).
(9) M. Heidelberger and C. F. C. MacPherson, Science, 97, 405 (1943); 98, 63 (1943).

#### Experimental

Antipneumococcal antibodies were produced by injection anti-SIII<sup>11</sup> N per ml. Specific precipitates of C-anti-C, SIII-anti-SIII and SII-anti-SII were obtained from about 100-ml. portions of serum, which had been in the refrigerator until the complement was destroyed, washed free from excess serum protein<sup>9,10,12</sup> dissolved in water with 0.5 ml. of 0.1 M NaOH, and made up to a known volume. Four

0.1 *M* NaOH, and made up to a known volume. Four samples of normal human gamma globulin were available for comparison with the human antibodies.<sup>13</sup> **Hexosamine/Total N Ratio.**—Aliquots of the dissolved SII-anti-SII and SIII-anti-SIII specific precipitates were analyzed for nitrogen by the Markham micro-Kjeldahl method<sup>14,12</sup> and for hexosamine by a modification<sup>15</sup> of the Elson-Morgan procedure.<sup>6</sup> The hexosamine values were corrected for the color given in this reaction by the SII and SIII in the dissolved precipitates: these samples of poly-SIII in the dissolved precipitates; these samples of poly-saccharide gave color values equivalent to 3.3 and 0.9% hexosamine, respectively. Two lots of normal human gamma globulin were analyzed for nitrogen and hexosamine. C-anti-C precipitates were not suitable for deter-mining the hexosamine/total N ratio since the C substance has a high (22%) hexosamine content. Methylpentose/Total N Ratio --SIII-anti-SIII and C-

anti-C specific precipitates and three gamma globulin samples were analyzed for methylpentose and nitrogen,14 and the values for the specific precipitates corrected for the methylpentose color given by these polysaccharides; SIII and C gave color values equivalent to 1.4 and 0.8% methyl-pentose, respectively. SII-anti-SII specific precipitates were not used in determining the methylpentose/N ratios

since the SII sample contained 40% of methylpentose. Folin-Ciocalteu Color Equivalent.--SII-anti-SII SIII-anti-SIII specific precipitates and two gamma globulin samples were used. SII and SIII contain no nitrogen and give no color with the Folin-Ciocalteu tyrosine reagent and no correction for their presence in the precipitates was necessary. Appropriate dilutions of known nitrogen content were analyzed as described by Heidelberger and Mac-Pherson.<sup>9,12</sup> Color development at 7500 Å. was proportional to N up to about  $25 \ \mu\text{g}$ . N. The values of hexosamine/N, methylpentose/N and mean

(10) M. Heidelberger, C. M. MacLeod, S. J. Kaiser and B. Robinson, J. Exp. Med., 83, 303 (1946).

(11) C denotes the group specific polysaccharide of pneumococcus and SII and SIII the type-specific capsular polysaccharides of types II and III pneumococci.

(12) E. A. Kabat and M. M. Mayer, "Experimental Immunochemistry." C. C. Thomas, Springfield, Ill., 1948,

(13) E. A. Kabat and J. P. Murray, J. Biol. Chem., 182, 251 (1950). (14) R. Markham, Biochem. J., 36, 790 (1942).

(15) K. Meyer, E. M. Smyth and J. W. Palmer, J. Biol. Chem., 119, 491 (1937).